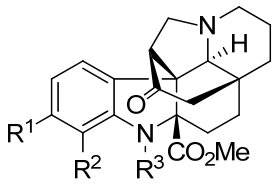
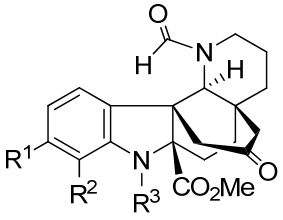
CHAPTER THREE

BIOMIMETIC ELECTROCHEMICALLY-MEDIATED PARTIAL SYNTHESIS OF DANUPHYLLINE B

3.1 Introduction

Danuphylline (**46**) was first isolated from the North Borneo species, *Kopsia dasyrachis*.[46,47] The structure of danuphylline represents a novel skeletal arrangement in which a new ring system has been formed as a result of cleavage of the C(5)-C(6) bond of the methyl chanofruticosinate precursor **44**, which was the predominant alkaloid present. Danuphylline (**46**) can thus be considered a "*seco*-methyl chanofruticosinate" and represents the first member of this group isolated as a natural product. A second danuphylline congener, danuphylline A (**47**) was subsequently isolated from the Chinese *K. arborea* (*K. officinalis*).[48] This was followed by the isolation of danuphylline B (**30**) from the Malayan *K. arborea*.[49]





25 $R^1 = R^2 = R^3 = H$
44 $R^1,R^2 = OCH_2O$, $R^3 = CO_2Me$
45 $R^1 = R^2 = H$, $R^3 = CO_2Me$

30 $R^1 = R^2 = R^3 = H$
46 $R^1,R^2 = OCH_2O$, $R^3 = CO_2Me$
47 $R^1 = R^2 = H$, $R^3 = CO_2Me$

3.1.1 Danuphylline B (30) - Isolation and Structure

Danuphylline B (**30**), the third member belonging to the danuphylline group of ring-opened alkaloids was obtained as a minor alkaloid from the leaf extract of *K. arborea*.[49]

Compound **30** was obtained as a colorless oil, with $[\alpha]_D$ +47 (*c* 0.13, $CHCl_3$). The UV spectrum showed absorption maxima at 212, 239, and 594 nm, indicating the presence of a dihydroindole chromophore, while the IR spectrum showed bands due to NH (3347 cm^{-1}), ketone/ester (1721 cm^{-1}), and formamide (1661 cm^{-1}) functions. The EIMS showed a molecular ion at *m*/*z* 368, with other significant fragment peaks at *m*/*z* 339 (M – CHO), 309 (M – CO_2Me), and 281 (M – CO_2Me – CH_2CH_2). HREIMS measurements (*m*/*z* 368.1735) gave the formula $C_{21}H_{24}N_2O_4$ (calc. 368.1736), which differs from danuphylline A (**47**) by 58 mass units,[48] suggesting the replacement of a CO_2Me group with a H-atom in **30**. Analysis of the NMR data for the non-indolic portion of **30** showed a close correspondence to those of **46** and **47**, suggesting departure from these two alkaloids in the indole moiety. The ^1H and ^{13}C NMR data of **30** (Table 3.1) are essentially the same as those of **47**, except for the absence of peaks associated with the carbamate group. Danuphylline B (**30**) is therefore the N-decarbomethoxy congener of danuphylline A (**47**). The ^1H and ^{13}C NMR spectra of **30** are shown in Fig. 3.1 and Fig. 3.2, respectively.

Table 3.1: ^{1}H and ^{13}C NMR Spectroscopic Data of Danuphylline B (**30**)[a]

Position	^{1}H	^{13}C
2	-	77.0
3	2.73 m	34.7
	4.58 dd (12, 9)	
5	6.62 s	165.7
6	2.70 d (17)	39.2
	2.85 d (17)	
7	-	53.9
8	-	128.8
9	6.86 m	123.9
10	6.84 m	119.9
11	7.18 ddd (8, 7, 2)	129.3
12	6.86 m	111.1
13	-	148.9
14	1.75 m	19.2
	1.96 m	
15	1.30 dt (14, 9)	29.8
	1.63 m	
16	-	207.9
17	2.45 d (20)	45.9
	2.72 d (20)	
18	2.18 m	26.7
	2.01 m	
19	1.65 m	39.0
	1.96 m	
20	-	34.0
21	3.68 s	59.7
CO_2*Me*	3.62 s	52.5
CO_2Me	-	174.2

[a]$CDCl_3$, 400 MHz

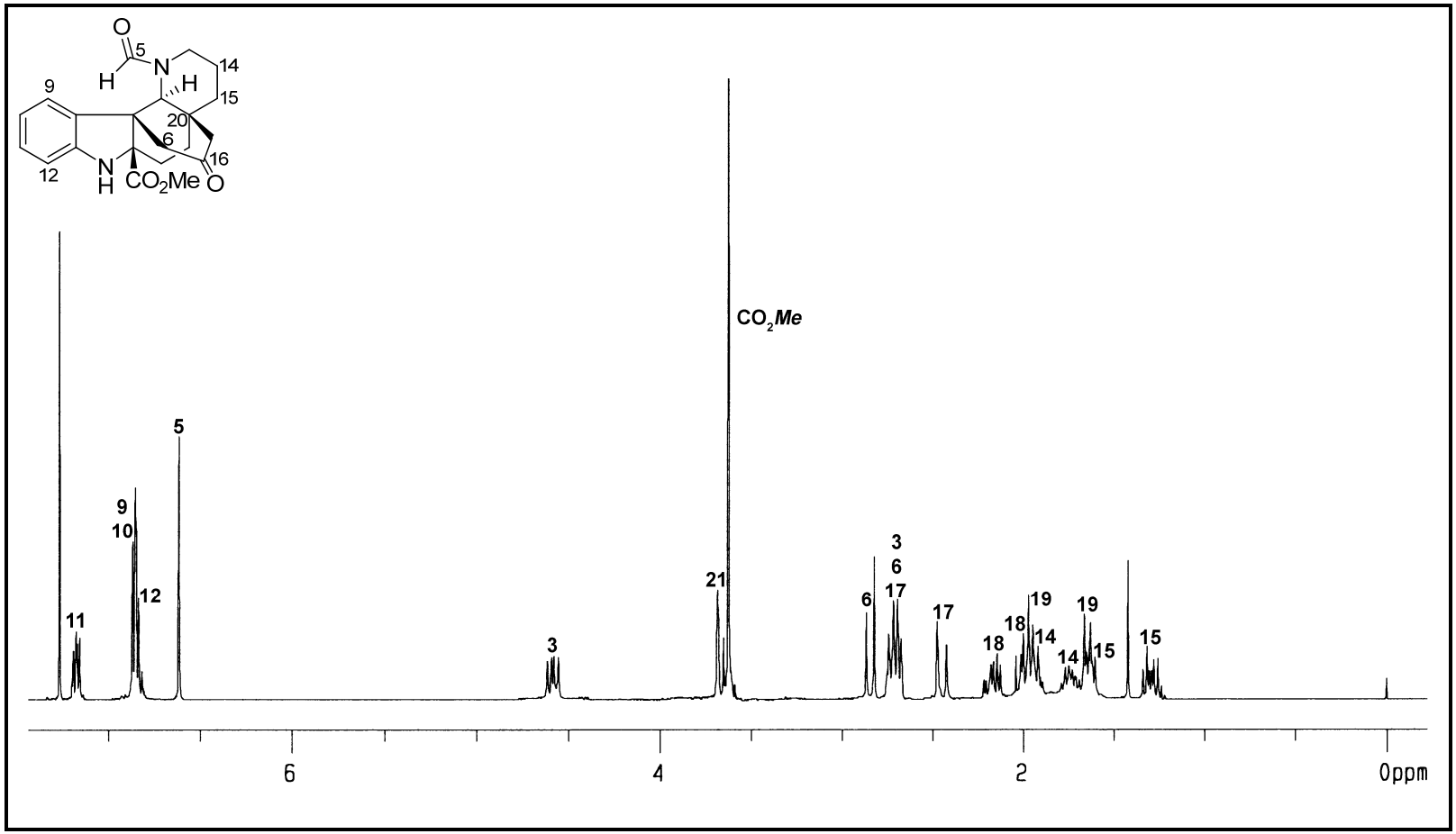


Fig. 3.1: ^{1}H NMR Spectrum ($CDCl_3$, 400 MHz) of Danuphylline B (**30**)

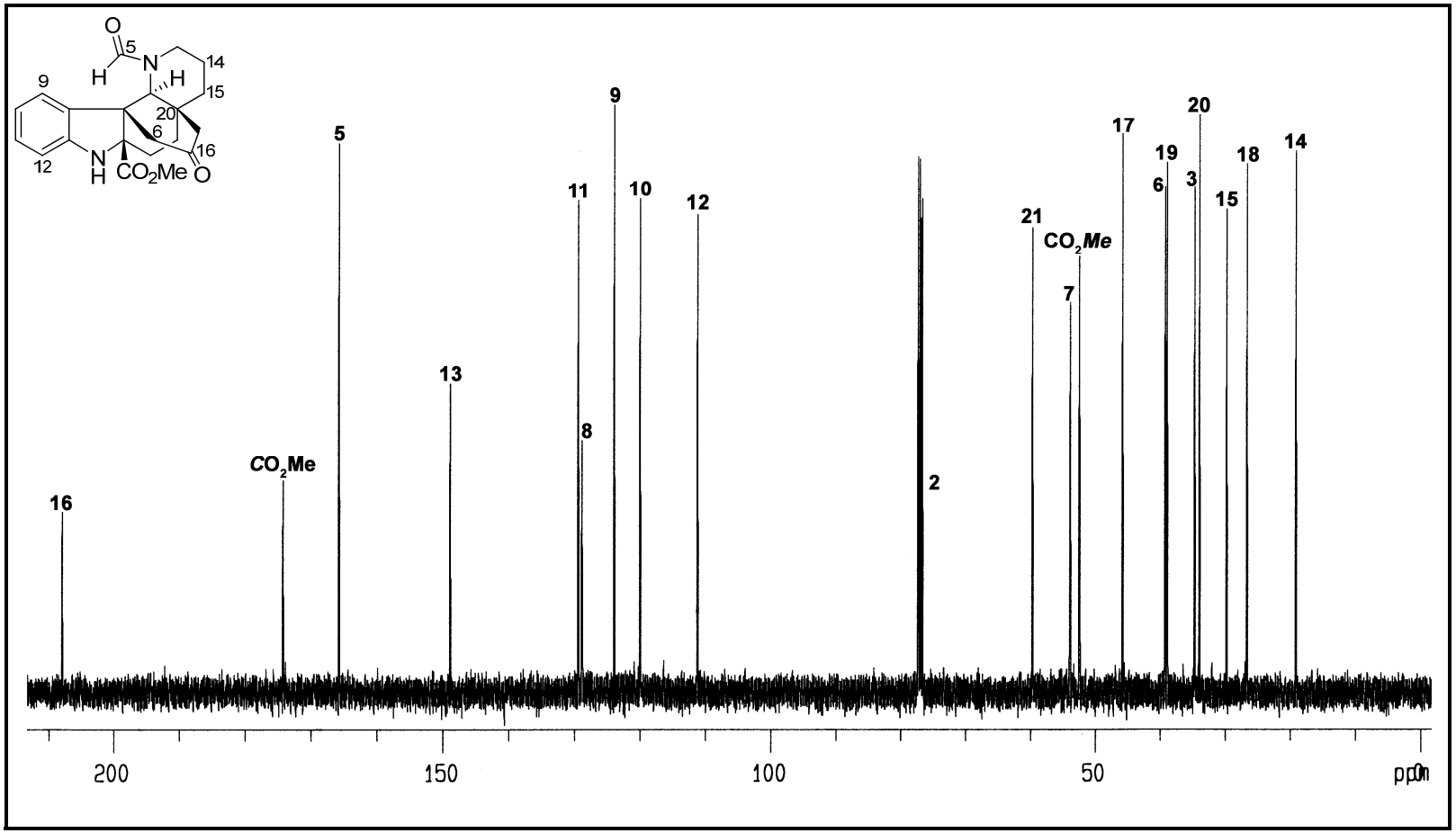
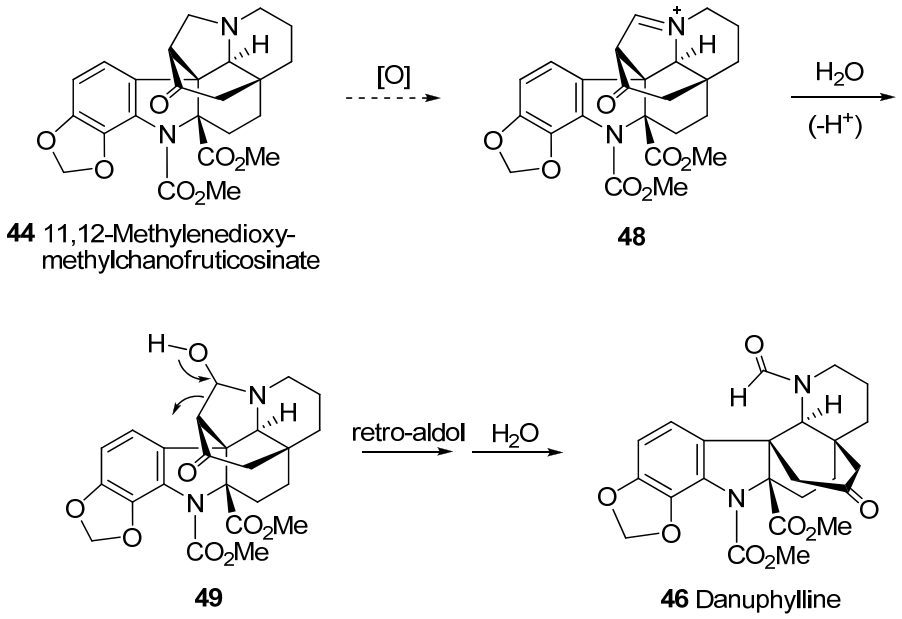


Fig. 3.2: ^{13}C NMR Spectrum ($CDCl_3$, 100 MHz) of Danuphylline B (**30**)

3.1.2 Proposed Biogenesis and Biomimetic Synthesis of Danuphylline (46)

It has been previously suggested that a possible origin of the danuphylline-type alkaloids is via a retro-aldol reaction of an unstable carbinol amine **49**, in turn derived from hydrolysis of the iminium ion **48**, formed from oxidation of the appropriate hexacyclic methyl chanofruticosinate precursor, **44** (Scheme 3.1).[46,47]



44 11,12-Methylenedioxy-methylchanofruticosinate

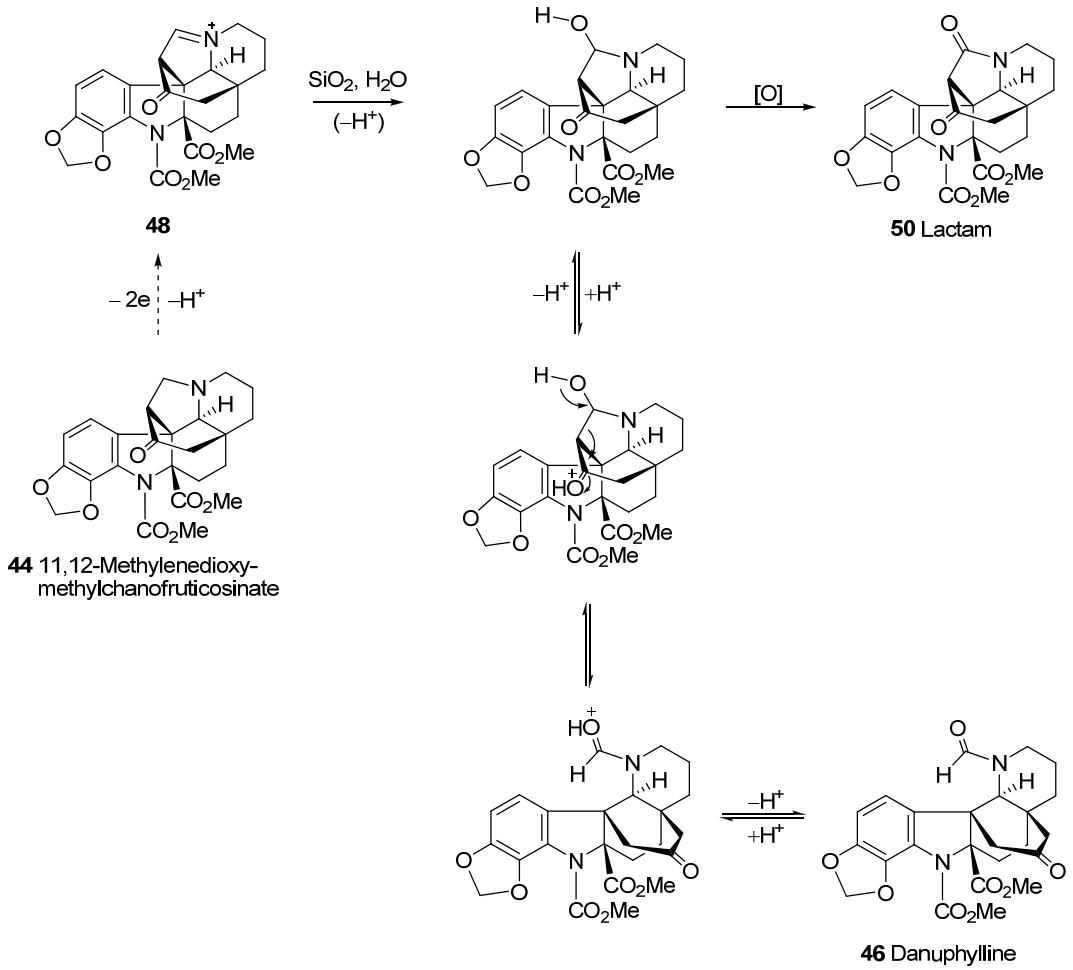
48

49

46 Danuphylline

Scheme 3.1

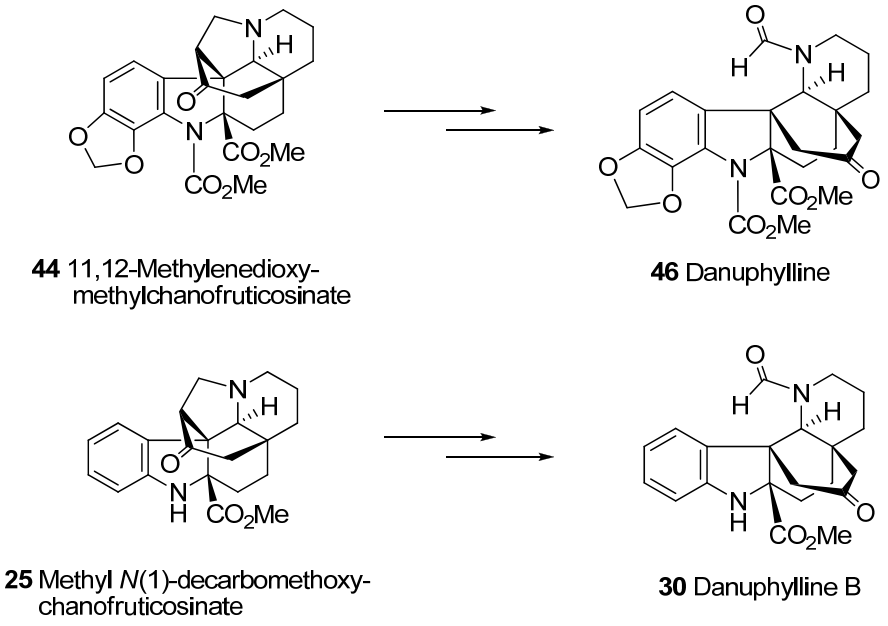
Based on the above proposal, an electrochemically-mediated partial synthesis of danuphylline (**46**) was implemented. The key step was the generation of the iminium ion **48** by anodic oxidation of the appropriate methyl chanofruticosinate precursor **44**, which on subsequent transformation during workup led to the desired ring-opened product. This approach yielded in addition to danuphylline (**46**), the lactam **50** as a minor side product (Scheme 3.2).[47]



Scheme 3.2

3.1.3 Proposed Biomimetic Partial Synthesis of Danuphylline B (30)

In view of the successful partial synthesis of danuphylline (**46**) via electrochemical oxidation of **44**,[47] it was envisaged that a similar biomimetic transformation of an appropriate methyl chanofruticosinate precursor such as **25** to danuphylline B (**30**) might also be feasible (Scheme 3.3). Furthermore, the alkaloid **25** was readily available as it constituted one of the many alkaloids obtained from the leaf extract of *K. arborea*.[50]



Scheme 3.3

Methyl *N*(1)-decarbomethoxychanofruticosinate (**25**) was obtained in moderate amount from the leaf extract of *K. arborea*,[50] and was also previously isolated from other *Kopsia* species.[51]

Compound **25** was obtained as a yellowish oil, with $[\alpha]_D$ +266 (*c* 0.25, $CHCl_3$). The UV spectrum (209, 234, and 293 nm) suggested the presence of a dihydroindole chromophore. The EIMS of **25** showed an M^+ at *m/z* 352, consistent with the molecular formula $C_{21}H_{24}N_2O_3$. In agreement with this, the ^{13}C NMR spectrum showed a total of 21 carbon resonances (one methyl, seven methylenes, six methines, and seven quaternary carbons). The ^1H NMR spectrum showed the presence of four aromatic hydrogens of an unsubstituted indole chromophore (δ_H 7.11, H-9; δ_H 6.79, H-10; δ_H 7.09, H-11; δ_H 6.76, H-12) and an indolic NH at δ_H 4.48. A conspicuous feature of the ^1H NMR spectrum of **25** is the H-6 resonance which is a doublet at δ_H 3.31 with J = 6 Hz, a feature which confirms the disposition of the N-4 lone pair which is directed outwards (*cis* to H-21). This conformation results in a dihedral angle of ca 90° between

H-6 and H-5β which accounts for the multiplicity of the H-6 signal observed in the ^{1}H NMR spectrum. The ^{1}H and ^{13}C NMR spectral data of **25** are summarized in Table 3.2 and the ^{1}H and ^{13}C NMR spectra of **25** are shown in Fig. 3.3 and Fig. 3.4, respectively.

Table 3.2: ^{1}H and ^{13}C NMR Spectroscopic Data of Compound **25**[a]

Position	^{1}H	^{13}C
2	-	73.8
3α	2.95 m	46.5
3β	2.95 m	
5α	2.95 m	52.6
5β	3.76 dd (11, 6)	
6	3.31 d (6)	55.1
7	-	57.6
8	-	133.1
9	7.11 d (7)	123.9
10	6.79 t (7)	119.8
11	7.09 t (7)	128.0
12	6.76 d (7)	110.1
13	-	147.7
14α	1.28 m	17.4
14β	1.85 m	
15α	1.28 m	34.9
15β	1.85 m	
16	-	209.3
17α	2.04 d (19)	42.5
17β	2.84 d (19)	
18α	1.70 m	27.4
18β	1.90 m	
19α	1.28 m	34.6
19β	1.54 br d (13)	
20	-	36.1
21	2.50 s	68.3
CO_2*Me*	3.61 s	52.2
CO_2Me	-	175.0

[a]$CDCl_3$, 400 MHz

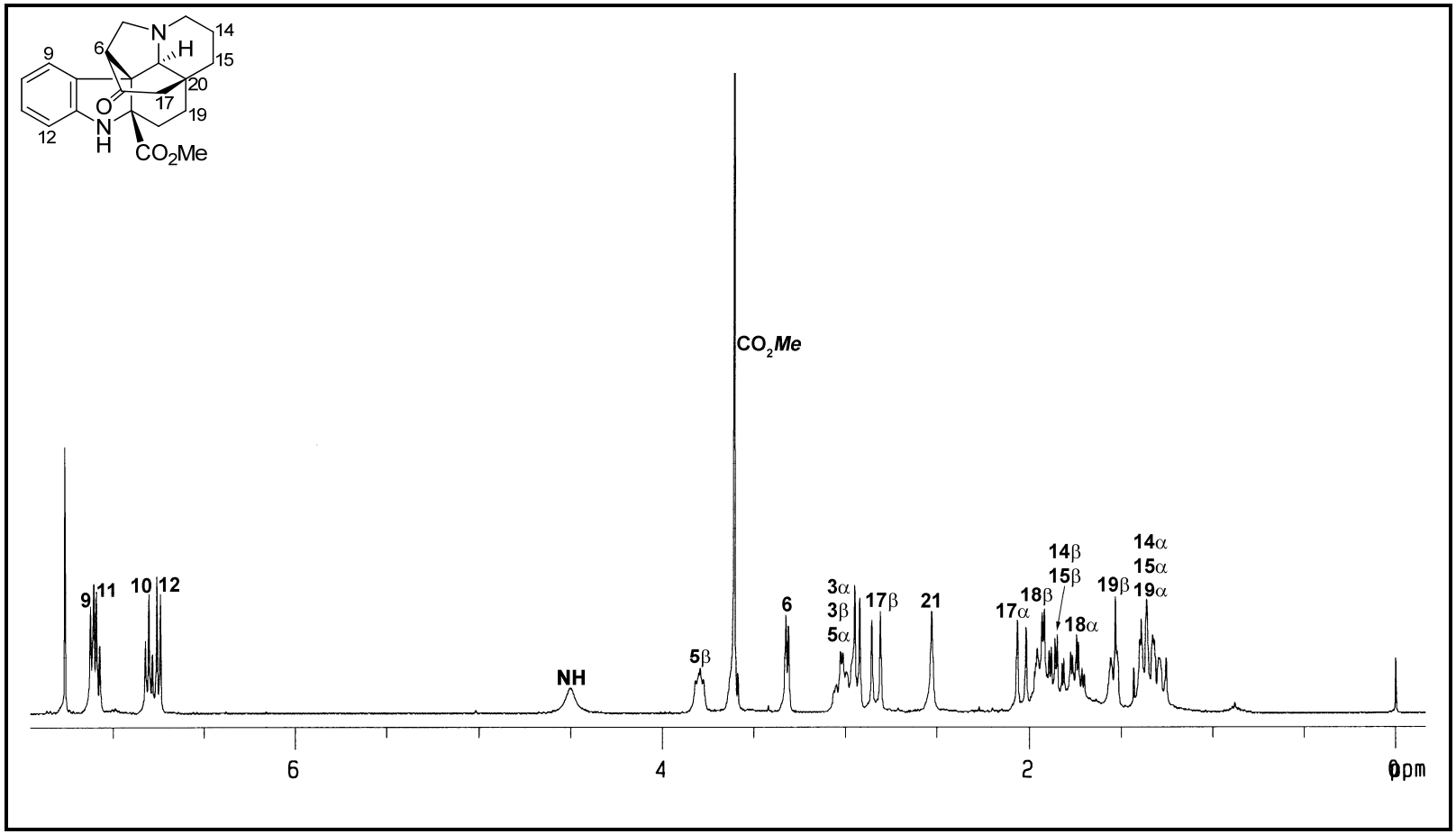


Fig. 3.3: ^{1}H NMR Spectrum ($CDCl_3$, 400 MHz) of Methyl *N*(1)-decarbomethoxychanofruticosinate (**25**)

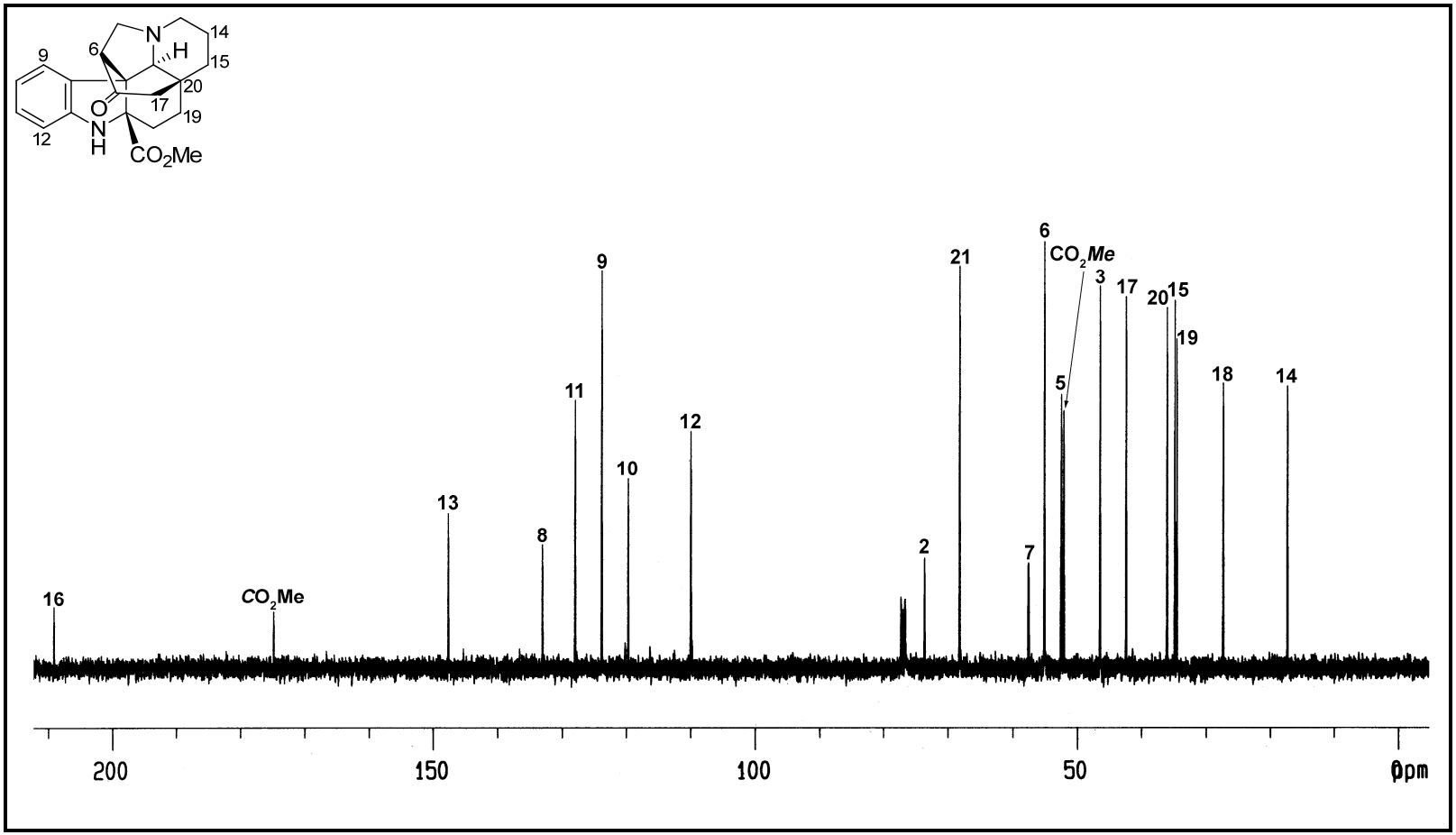


Fig. 3.4: ^{13}C NMR Spectrum ($CDCl_3$, 100 MHz) of Methyl *N*(1)-decarbomethoxychanofruticosinate (**25**)

3.2 Results and Discussion

3.2.1 Direct Anodic Oxidation of Methyl *N*(1)-decarbomethoxychano-fruticosinate (25)

Anodic oxidation of **25** (Pt anode, 30% CH_2Cl_2-MeCN, 0.1 M Et_4NClO_4) showed two irreversible waves at 1.03 and 1.23 V versus Ag/AgCl in the potential range studied, as revealed by cyclic voltammetry (Fig. 3.5). Controlled potential electrolysis (Pt gauze anode, Pt cathode) of **25** at the first potential peak (1.2 V) in the presence of 2,6-lutidine as proton scavenger, was allowed to proceed until consumption of 2 $Fmol^{-1}$ of charge. In the course of the electrooxidation, the originally colorless solution changed to a deep orange-brown coloration. Removal of the solvent in vacuo followed by removal of the bulk of the supporting electrolyte by precipitation with CH_2Cl_2 yielded a dark brown mixture. TLC of the crude product mixture did not show formation of any significant product.

The failure to obtain any significant product from the electrooxidation can be attributed to the presence of the indolic NH in **25**, which may interfere in some way during the electrochemical process, a supposition supported by our previous experience of electrochemical oxidation of indole derivatives.[46,52–54]

We therefore proceeded to protect the indolic NH of **25** prior to carrying out the electrochemical oxidation (Scheme 3.4).

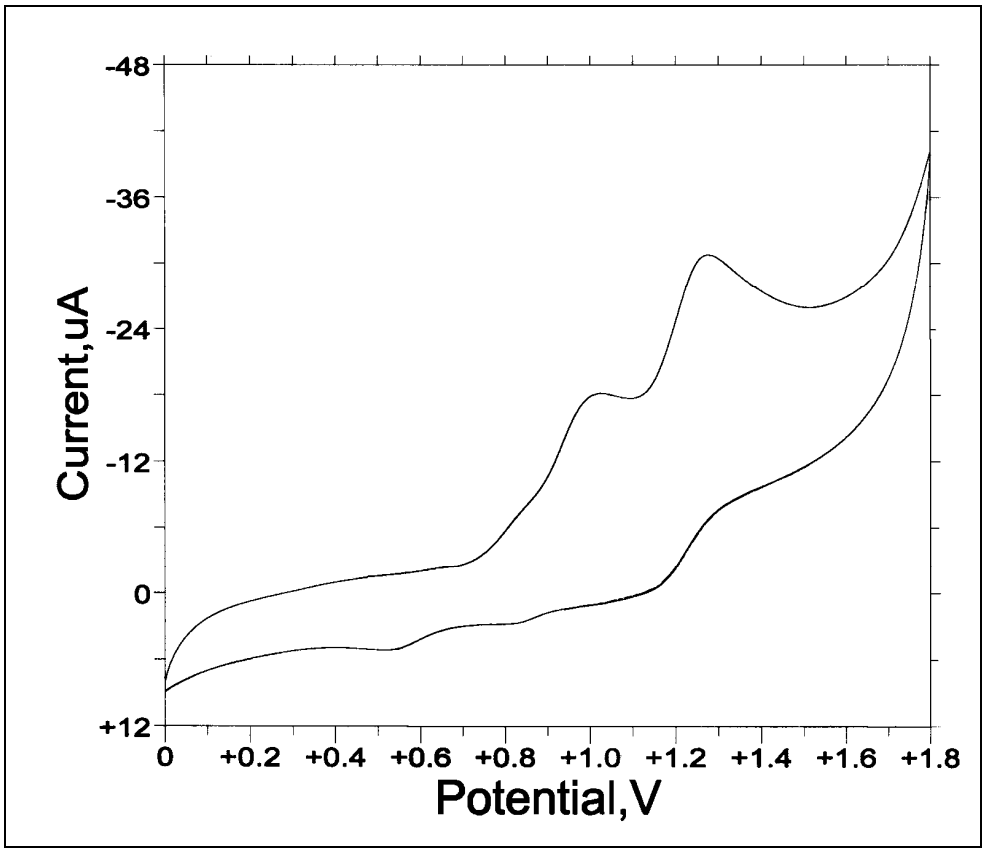
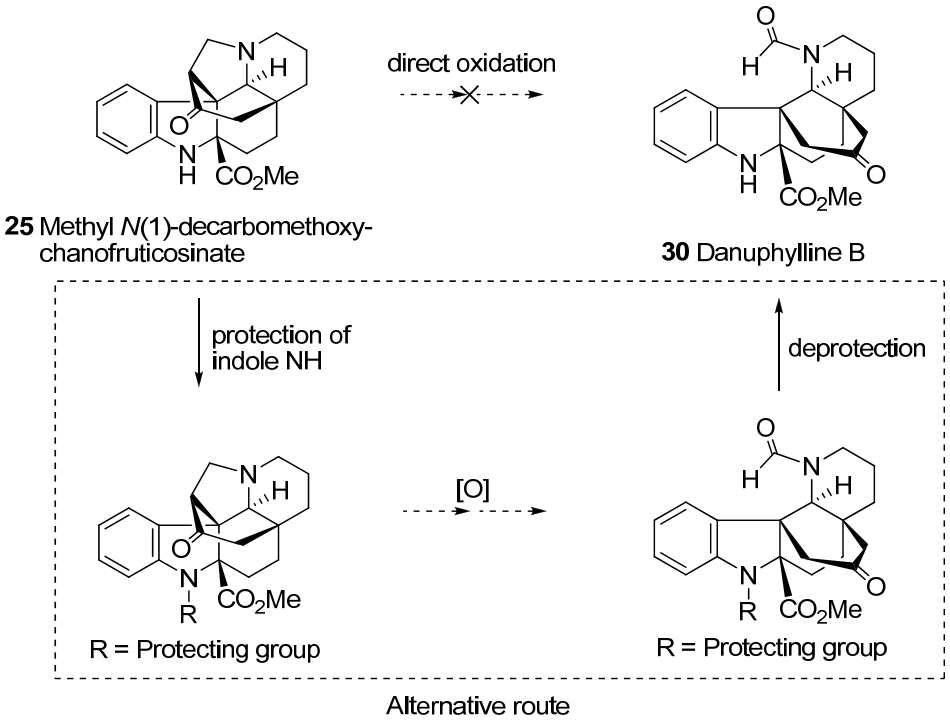


Fig. 3.5: Cyclic voltammogram of **25** at Pt electrode in 30% CH_2Cl_2-MeCN containing Et_4NClO_4 (0.1 M). E/V versus Ag/AgCl, sweep rate = 100 mV/s



Scheme 3.4

3.2.2 Preparation of N-Boc Protected Derivative of 25

An indolic NH can be protected as the carbamate, amide, imine, enamine, or alkyl derivative. A methyl carbamate group was initially chosen since a successful electrochemically-mediated transformation would give danuphylline A (**47**), which on N-1 deprotection would in turn yield danuphylline B (**30**).

Unfortunately, attempted installation of a methyl carbamate group on **25** with methyl chloroformate under various conditions proved problematic, and in view of the limited amount of **25** available, preparation of the N-Boc derivative of **25** was next attempted.

Initial experiments employing mild reaction conditions [e.g., di-*tert*-butyl dicarbonate (Boc_2O), in the presence of triethylamine (TEA) in MeCN at rt] did not give the desired product.[55] Attempts to vary the reaction conditions, such as raising the temperature of the reaction, changing the solvent, or changing the base, were also unsuccessful. Harsher reaction conditions were therefore required to install the Boc protecting group.[56]

Thus, initial premixing of the methyl chanofruticosinate **25** with 2.2 equivalent of Boc_2O, was followed by addition of sodium bis(trimethylsilyl)amide (NaHMDS, 2.2 equiv) following the method of Overman and coworkers.[56] This however gave rise to a mixture of the desired N-Boc protected derivative **51**, accompanied by the doubly-acylated enol carbonate **52**, with the latter obtained as the major product (e.g., ratio of **51** : **52**, ca. 1 : 2.4).

The N-Boc protected derivative **51** was obtained as a colorless oil with $[\alpha]_D$ +106 (*c* 0.65, $CHCl_3$). The IR spectrum showed bands due to various carbonyl functions (1744 and 1711 cm^{-1}, broad). A notable difference in the ^1H NMR spectrum of **51** when compared with that of **25** is the absence of the characteristic indolic NH signal and the

presence of the characteristic singlet of the *t*-butylcarbamate group (δ_H 1.5). The ^{13}C NMR spectrum (Table 3.3) also showed additional resonances due to the *t*-butyl group (δ_C 28.2 and 82.4) associated with the carbamate carbonyl signal (δ_C 152.4). The mass spectrum showed a molecular ion at *m/z* 452, which is 100 mass units higher than that of **25**, while HREIMS measurements gave the expected molecular formula $C_{26}H_{32}N_2O_5$. The ^1H and ^{13}C NMR spectral data of **51** are summarized in Table 3.3, and the ^1H and ^{13}C NMR spectra of **51** are shown in Fig. 3.6 and Fig. 3.7, respectively.

The doubly-acylated enol carbonate **52** was obtained as a light yellowish oil with $[\alpha]_D$ +99 (*c* 0.71, $CHCl_3$). The IR spectrum showed bands due to various carbonyl functions (1811, 1752, and 1708 cm^{-1}). Notable differences in the ^1H NMR spectrum of **52** when compared with that of **25** include the absence of the characteristic indolic NH signal as well as the AB doublets due to the isolated methylene unit at C-17, and the presence of an additional olefinic proton at δ_H 4.82, attributed to the enolic H-17, in addition to the signals due to the two *t*-butyl groups of the carbamate/carbonate functions (δ_H 1.52, 1.47). The ^{13}C NMR of **52** showed the absence of the characteristic carbonyl signal of C-16, but displayed additional resonances at δ_C 151.2 and 120.7, attributed to the olefinic C-16 and C-17, respectively, as well as two sets of the characteristic *t*-butoxylcarbonyl group signals associated with the carbamate (δ_C 82.6, 27.7, 150.1) and enol carbonate (δ_C 82.0, 28.1, 150.1) functions. The EIMS of **52** showed an M^+ at *m/z* 552, which is 200 mass units higher than that of **25**. HREIMS measurements gave the molecular formula $C_{31}H_{40}N_2O_7$ consistent with the addition of two *t*-butoxylcarbonyl groups in **52** when compared to that of **25**. The ^1H and ^{13}C NMR spectral data of **52** are summarized in Table 3.3, and the ^1H and ^{13}C NMR spectra of **52** are shown in Fig. 3.8 and Fig. 3.9, respectively.

Table 3.3: ^{1}H and ^{13}C NMR Spectroscopic Data of N-Boc Protected Derivative **51** and Doubly-acylated Enol Carbonate **52**[a]

Position	**51**		**52**	
	^{1}H	^{13}C	^{1}H	^{13}C
2	-	73.2	-	73.0
3	2.97 d (6)	46.4	2.92 t (14)	48.0
	2.99 m		2.99 t (14)	
5	2.78 d (11.5)	52.1	3.21 d (10)	57.9
	3.65 dd (11.5, 6.5)		3.66 dd (10, 5)	
6	2.82 d (6)	56.9	2.56 d (5)	47.2
7	-	57.6	-	60.9
8	-	133.4	-	132.8
9	7.32 dd (8, 1)	124.9	7.67 d (8)	126.0
10	6.96 td (8, 1)	122.6	7.01 t (8)	122.9
11	7.21 td (8 ,1)	128.4	7.24 t (8)	128.4
12	7.78 br d (8)	114.1	7.88 br s	114.5
13	-	141.6	-	140.9
14	1.44 m	17.3	1.71 m	19.2
	1.61 m		1.25 m	
15	1.33 m	35.6	1.32 m	37.5
	1.65 m		1.82 m	
16	-	208.2	-	151.2
17	2.15 d (19.5)	43.7	4.82 s	120.7
	2.71 d (19.5)			
18	2.41 m	25.1	1.71 m	26.5
	2.58 m		2.77 dd (13.5, 8)	
19	1.31 m	34.4	1.39 m	31.6
	1.83 m		1.86 m	
20	-	36.3	-	36.7
21	2.50 s	67.6	2.73 s	64.5
CO_2Me	3.57 s	52.0	3.74 s	51.5
NCO_2CMe_3	1.50 s	28.2	1.52 s	28.1
OCO_2CMe_3	-	-	1.47 s	27.7
NCO_2CMe_3	-	82.4	-	82.6
OCO_2CMe_3	-	-	-	82.0
CO_2Me	-	171.1	-	171.4
NCO_2CMe_3	-	152.4	-	150.1
OCO_2Me_3	-	-	-	150.1

[a]$CDCl_3$, 400 MHz

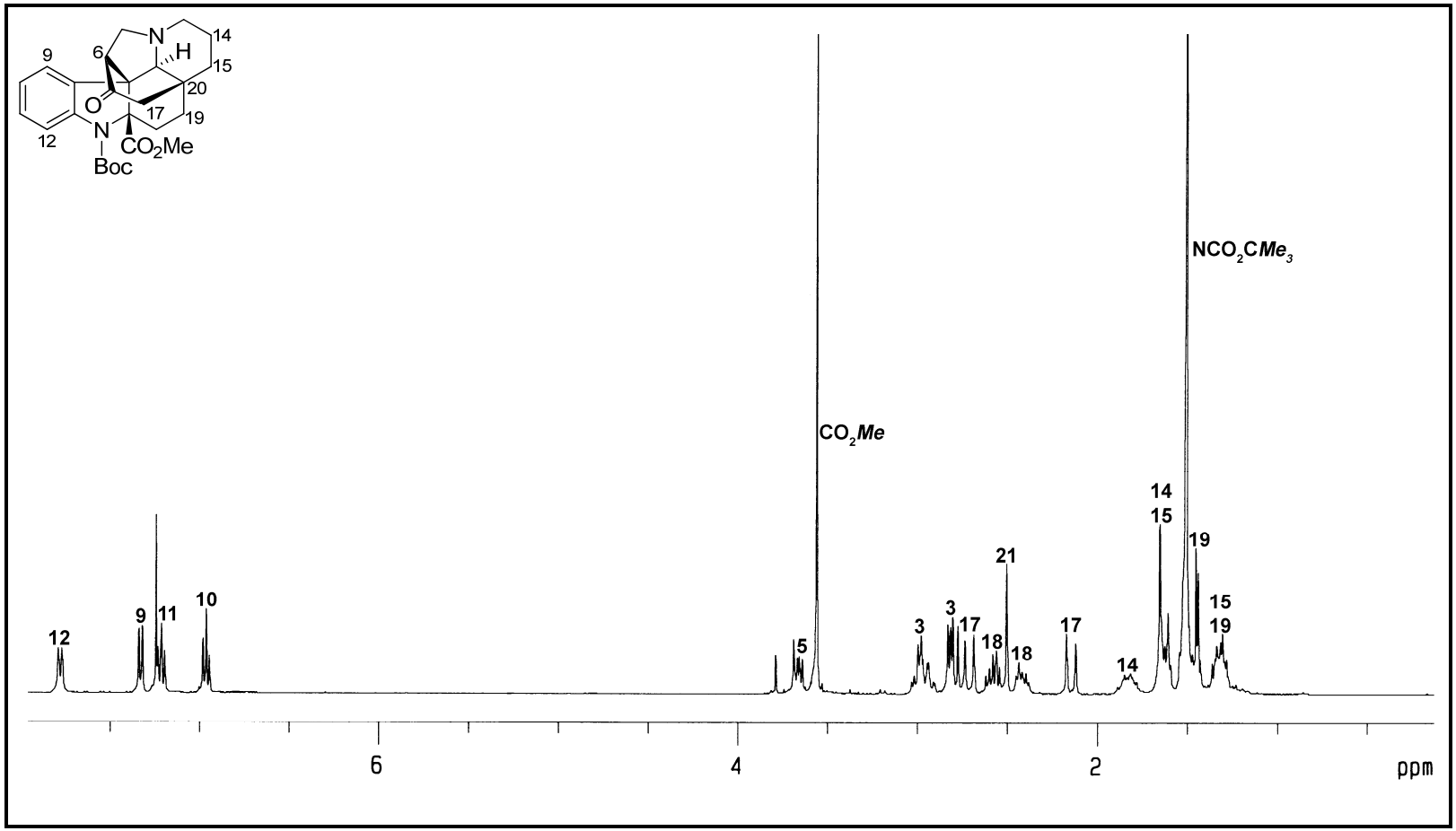


Fig. 3.6: ^{1}H NMR Spectrum ($CDCl_3$, 400 MHz) of Compound **51**

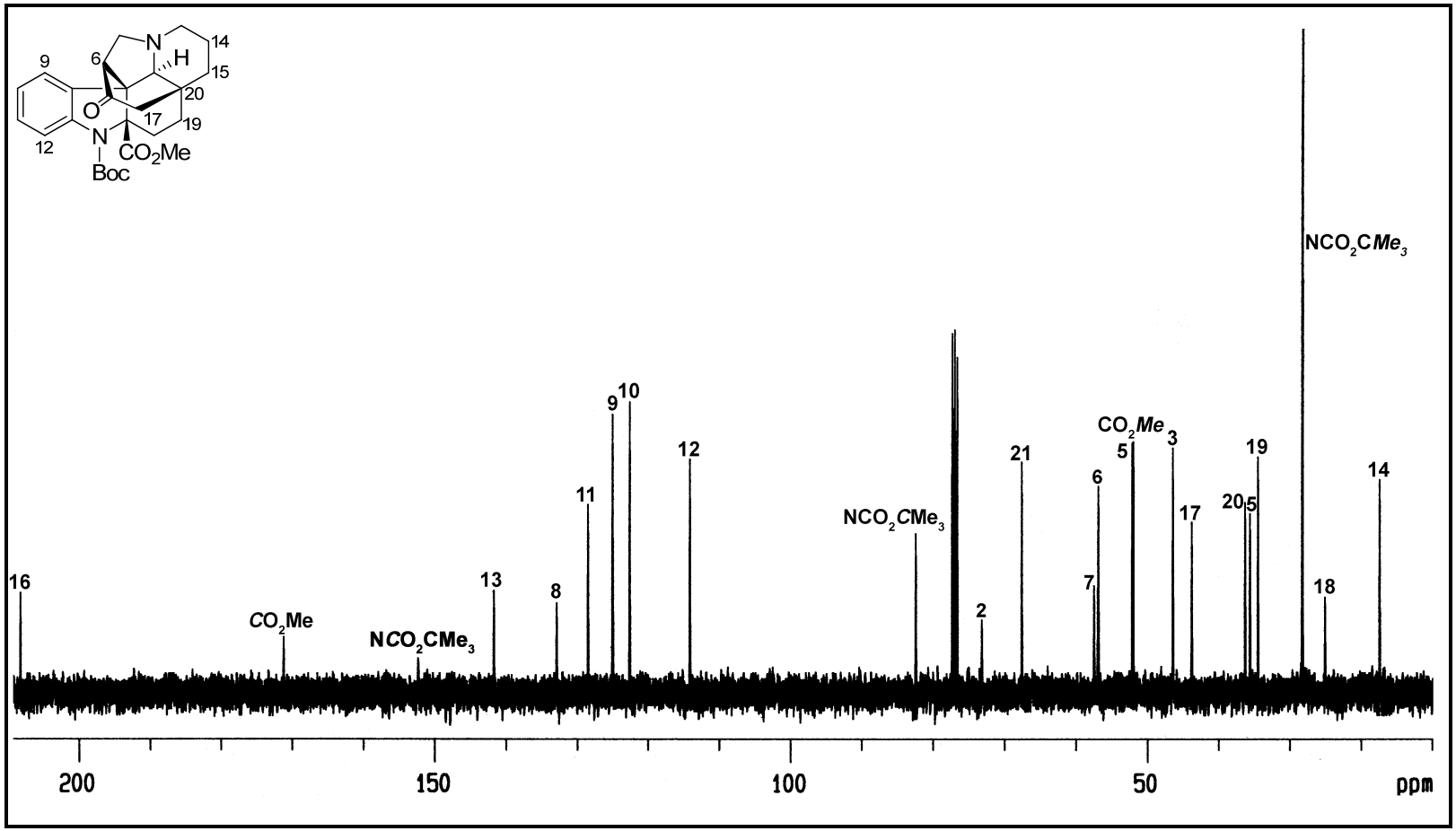


Fig. 3.7: ^{13}C NMR Spectrum ($CDCl_3$, 100 MHz) of Compound **51**

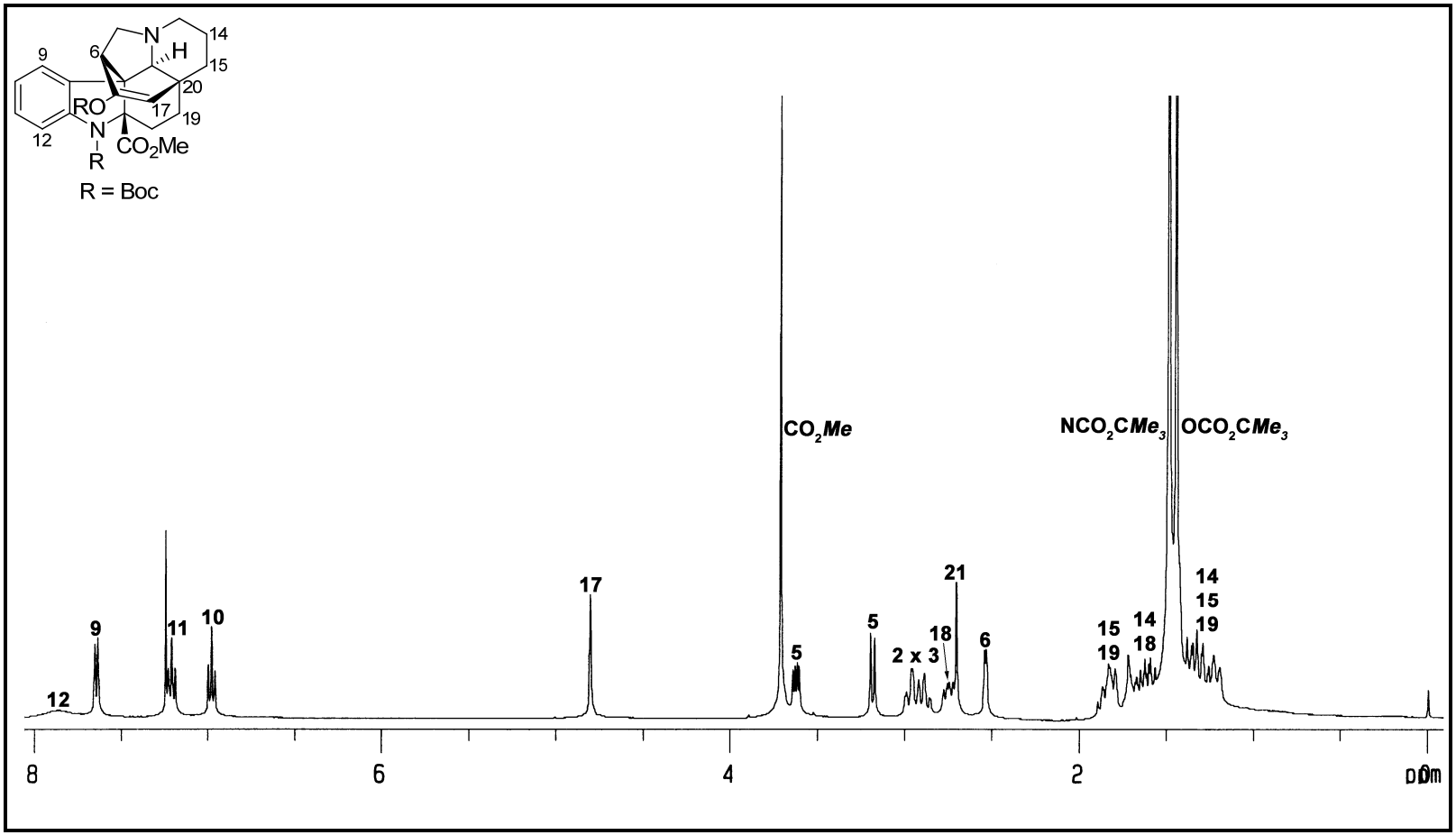


Fig. 3.8: ^{1}H NMR Spectrum ($CDCl_3$, 400 MHz) of Compound **52**

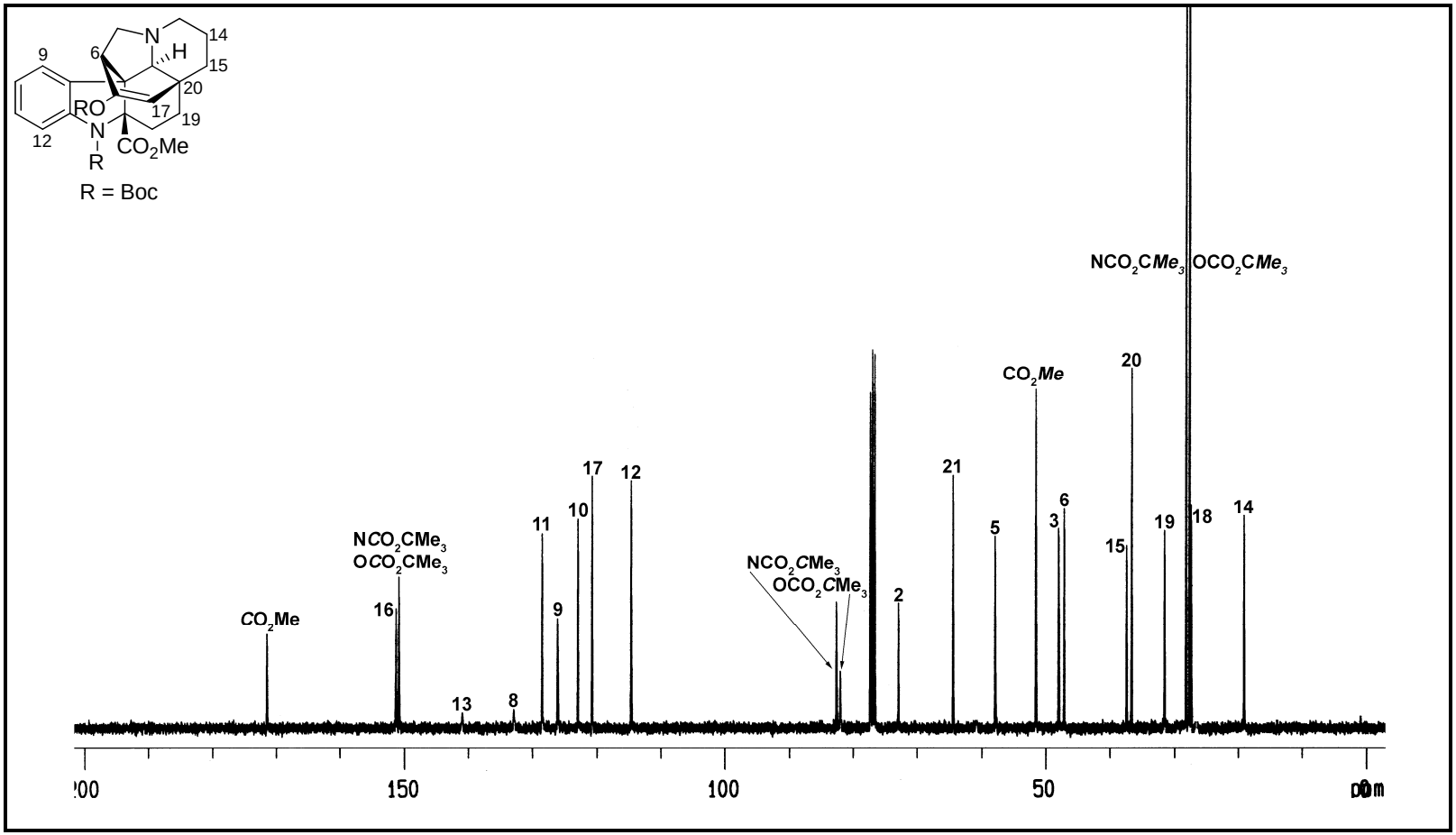
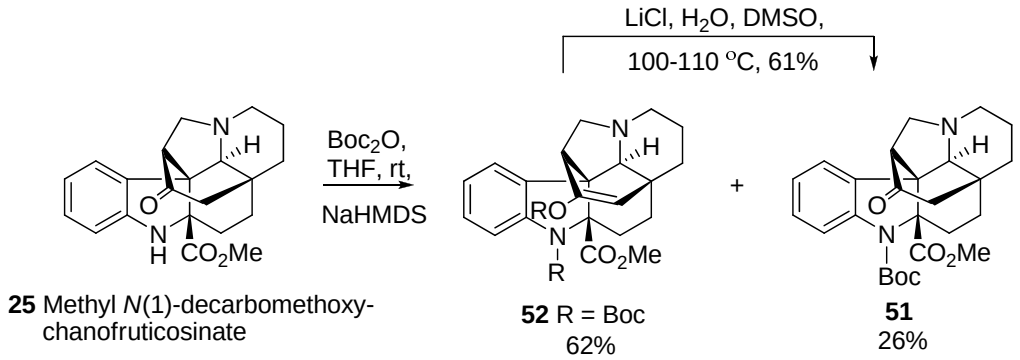


Fig. 3.9: ^{13}C NMR Spectrum ($CDCl_3$, 100 MHz) of Compound **52**

Attempts to protect the C-16 carbonyl function prior to the protection of the indolic NH via ketal formation[57] using various methods were not successful, as were atttempts to increase the yield of the **51** over **52** (through varying the ratio of the reagents, changing the order of addition of the reagents, or employing lower temperatures). Fortunately, it was found that **52** could be smoothly converted to the desired **51** via LiCl/DMSO-mediated decarboxylation (Krapcho decarboxylation) (Scheme 3.5).[58]



Scheme 3.5

3.2.3 Anodic Oxidation of Compound 51 and Subsequent Transformation to Danuphylline B (30)

Anodic oxidation of **51** (Pt anode, 30% CH_2Cl_2-MeCN, 0.1 M Et_4NClO_4) showed two irreversible waves at 1.07 and 1.75 V versus Ag/AgCl in the potential range studied (Fig. 3.10). Controlled potential electrolysis (Pt gauze anode, Pt cathode) of **51** at the first potential peak (1.2 V) in the presence of 2,6-lutidine was allowed to proceed until consumption of 2 $Fmol^{-1}$ of charge. This time the solution remained virtually colorless during the electrooxidation. TLC analysis of the electrolyzed solution (solvent CH_2Cl_2) showed the presence of a strong polar component near the baseline which is due to the iminium salt. Development of the TLC (SiO_2, 2% MeOH-CH_2Cl_2)

resulted in the formation of a new non-polar product (Boc-protected danuphylline B, **53**) at the expense of the original base-line product. Removal of the solvent in vacuo followed by removal of the bulk of the supporting electrolyte by precipitation with CH_2Cl_2, followed in turn by centrifugal preparative TLC (silica gel, CH_2Cl_2), resulted in the isolation of **53** (30%) (Scheme 3.6).

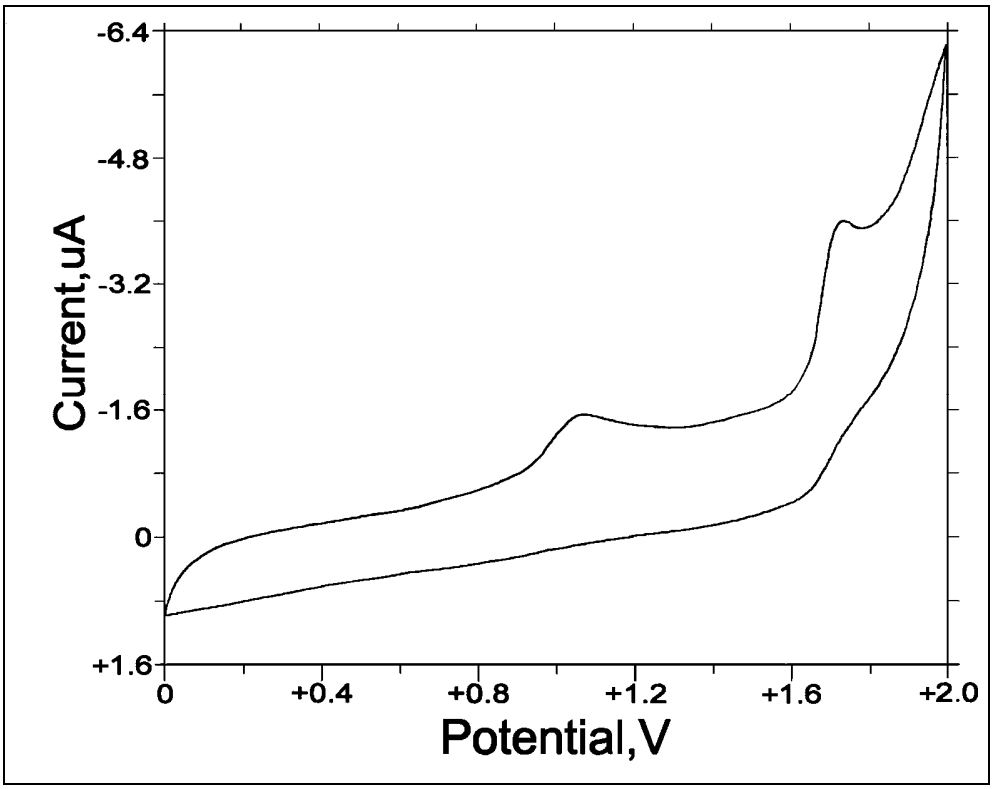
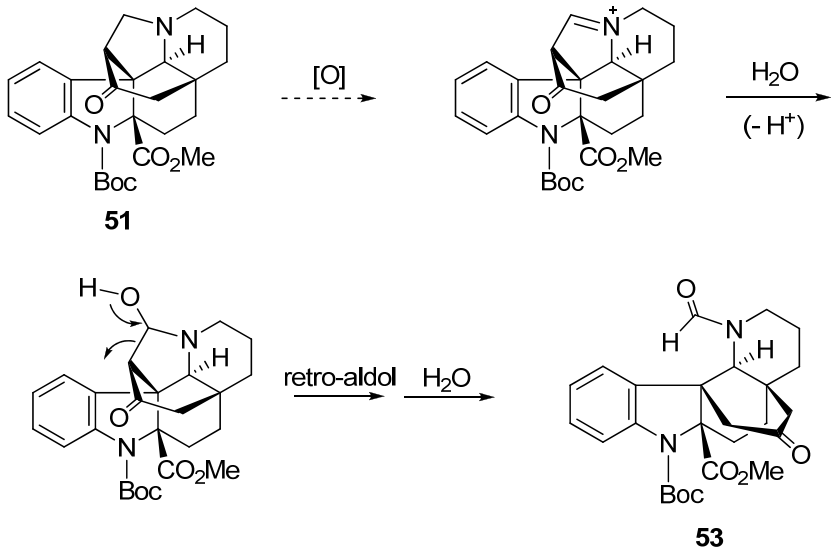


Fig. 3.10: Cyclic voltammogram of **51** at Pt electrode in 30% CH_2Cl_2-MeCN containing Et_4NClO_4 (0.1 M). E/V versus Ag/AgCl, sweep rate = 100 mV/s

The Boc-protected danuphylline B **53** was obtained as a colorless oil with $[\alpha]_D$ +28 (*c* 0.39, $CHCl_3$). The IR spectrum showed bands due to various carbonyl functions (1738, 1712, and 1673 cm^{-1}, broad). The EIMS of **53** showed an M^+ at *m/z* 468, differing from danuphylline B (**30**) by 100 mass units. HREIMS measurements gave the formula $C_{26}H_{32}N_2O_6$ (found *m/z* 468.2260, calcd 468.2260). The ^1H and ^{13}C NMR

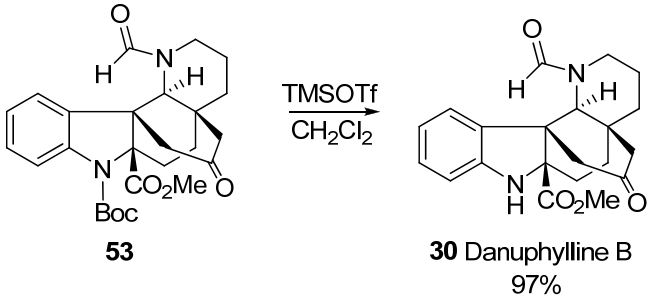
spectral data (Table 3.4) of **53** are virtually the same as those of **30**, except for the presence of signals due to a *t*-butoxylcarbamate group (δ_C 28.4, 83.1 and 153.6). The ^1H and ^{13}C NMR spectra of **53** are shown in Fig. 3.11 and Fig. 3.12, respectively.



Scheme 3.6

Finally, exposure of the Boc-protected danuphylline B **53** to trimethylsilyl trifluoromethanesulfonate (TMSOTf) in CH_2Cl_2 resulted in cleavage of the *t*-butoxylcarbonyl group to afford danuphylline B (**30**) in 97% yield (Scheme 3.7).

The spectroscopic data (^1H and ^{13}C NMR, IR, UV) and other analytical properties ($[\alpha]_D$ and R_f of TLC in different solvent systems) of synthetic danuphylline B (**30**) were indistinguishable from those of the natural danuphylline B (**30**).



Scheme 3.7

Table 3.4: ^{1}H and ^{13}C NMR Spectroscopic Data of Boc-protected Danuphylline B **53**[a]

Position	^{1}H	^{13}C
2	-	78.6
3	2.72 m	35.0
	4.52 dd (14, 9)	
5	6.38 s	165.8
6	2.37 d (17)	39.4
	2.83 d (17)	
7	-	54.1
8	-	129.2
9	7.31 t (7.5)	129.8
10	6.88 d (7.5)	123.8
11	7.04 t (7.5)	122.8
12	7.81 br d (7.5)	115.5
13	-	143.7
14	1.76 m	19.4
	1.89 m	
15	1.28 m	29.9
	1.71 m	
16	-	207.0
17	2.50 d (20)	46.1
	2.71 d (20)	
18	2.54 m	23.7
	3.20 br d (16.5)	
19	1.70 m	39.4
20	-	34.4
21	3.34 s	62.2
$CO_2\mathit{Me}$	3.57 s	52.7
$NCO_2C\mathit{Me}_3$	1.58 s	28.4
$NCO_2\mathit{C}Me_3$	-	83.1
$\mathit{C}O_2Me$	-	170.7
$N\mathit{C}O_2CMe_3$	-	153.6

[a]$CDCl_3$, 400 MHz

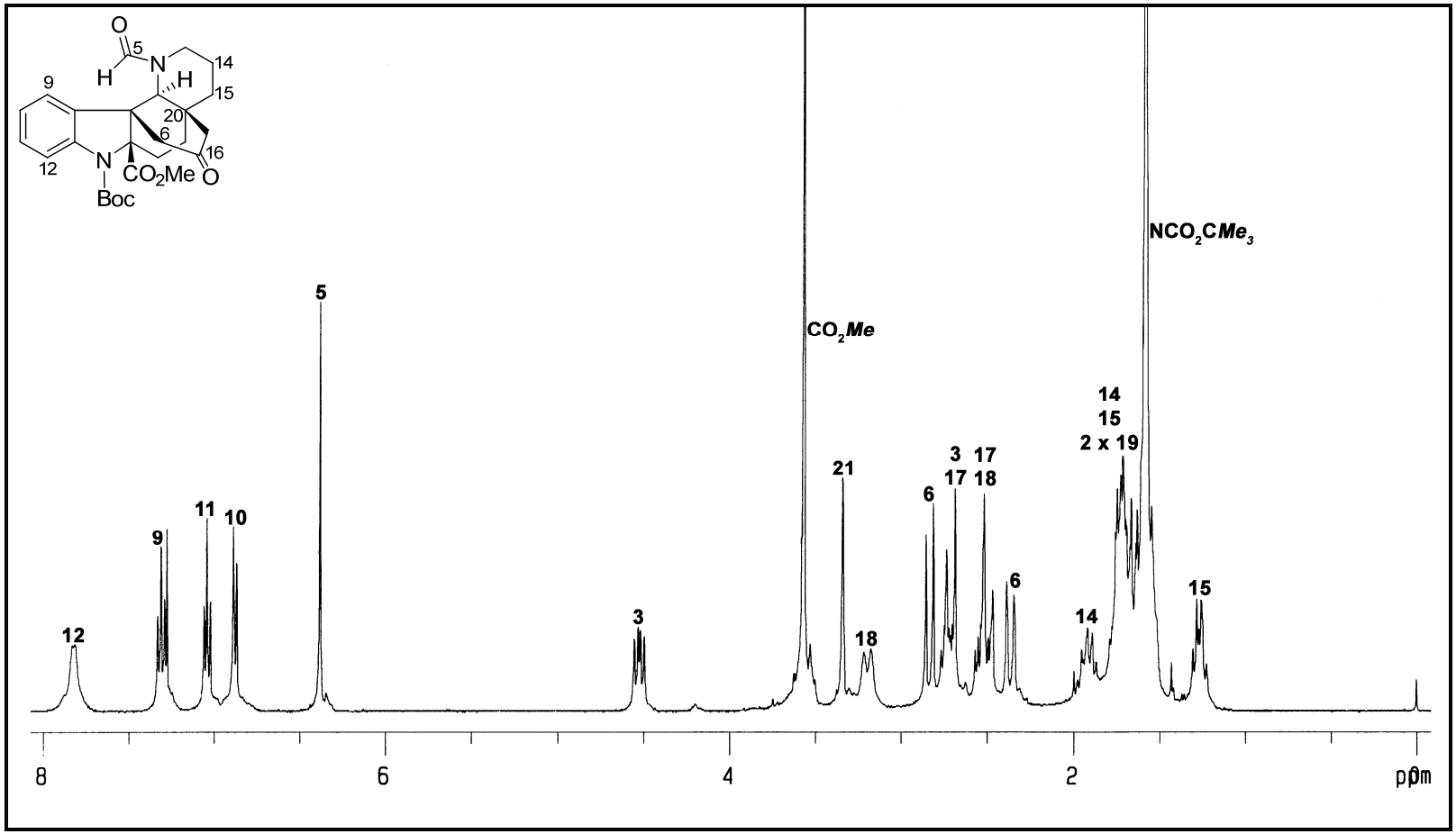


Fig. 3.11: ^{1}H NMR Spectrum ($CDCl_3$, 400 MHz) of Compound **53**

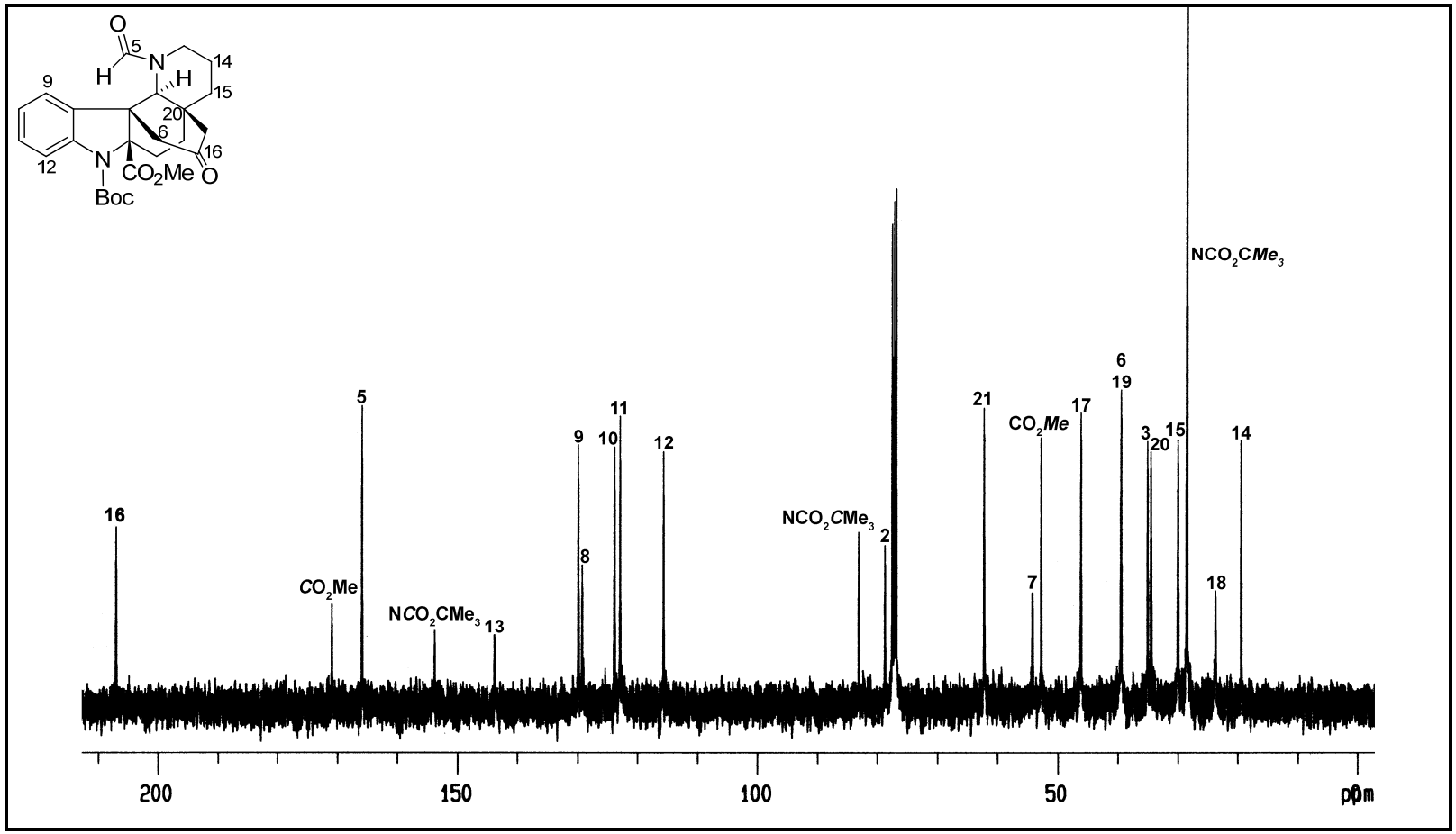


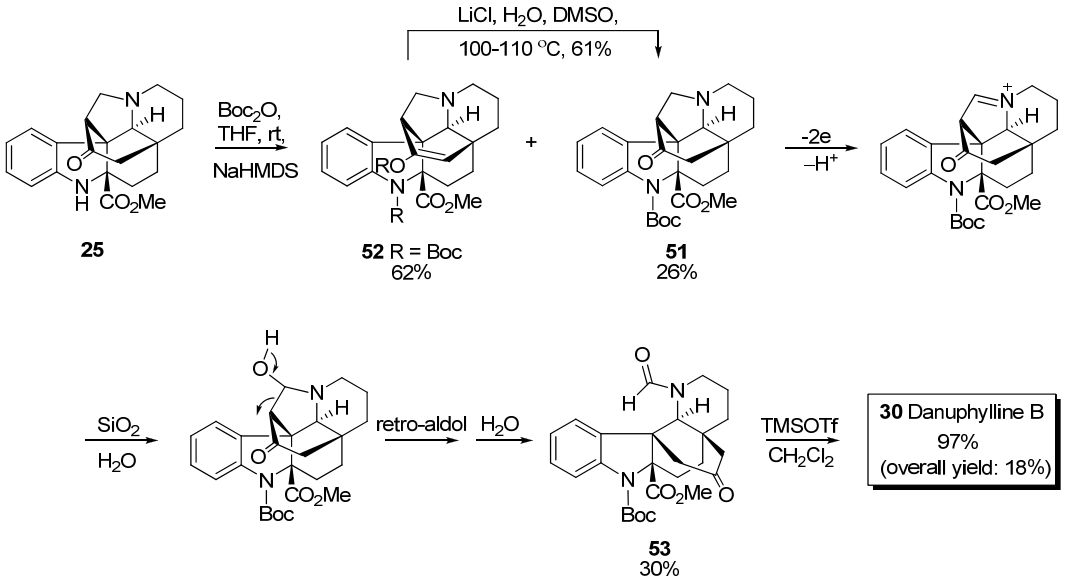
Fig. 3.12: ^{13}C NMR Spectrum ($CDCl_3$, 100 MHz) of Compound **53**

3.3 Conclusion

A biomimetic electrochemically-mediated semisynthesis of danuphylline B (**30**) has been achieved via anodic oxidation of the N(1)-Boc-protected methyl chanofruticosinate derivative **51**, followed in succession by passage of the iminium salt obtained through SiO_2 (CH_2Cl_2; 2% MeOH-CH_2Cl_2), and finally, deprotection by exposure to TMSOTf.

Installation of the Boc protecting group on the methyl chanofruticosinate precursor **25** gave both the desired N(1)-Boc derivative **51** as well as the doubly-acylated enol carbonate **52** which could however be smoothly transformed to **51** by Krapcho decarboxylation.

A summary of this transformation is shown in Scheme 3.8.



Scheme 3.8